REMARKS

Reconsideration of the present application in view of the above amendments and the following remarks is respectfully requested. Claims 1, 3-12, 15-18, 21, 39, 40, and 42-101 were pending. All of these claims have been canceled without acquiescence to the rejections in the Action or prejudice to future prosecution in a related application. New claims 102-124 have been added. Accordingly, claims 102-124 are pending. Claim 102 primarily correlates to previously pending claim 1. Additional support for this claim may be found, for example, in paragraphs [0006], [0008], [0044], and [0051] of the publication of the present application (*i.e.*, Publication No. US 2001/0031467). Support for claims 103 and 104 may be found, for example, in paragraph [0008]. Support for claims 105 and 106 may be found, for example, in paragraph [0103]. Support for claims 107 and 108 may be found, for example, in paragraph [0089]. Support for claim 109 may be found, for example, in paragraph [0090]. Support for claim 110 may be found, for example, in paragraph [0017]. Support for claim 111 may be found, for example, in paragraph [0022]. Support for claim 112 may be found, for example, in paragraph [0051] and Figure 4. Support for claim 113 may be found, for example, in paragraph [0036] and Figure 6. Support for claim 114 may be found, for example, in paragraphs [0006], [0022], [0036], and [0044]. Support for claim 115 may be found, for example, in original claim 17. Support for claim 116 may be found, for example, in paragraph [0100]. Support for claim 117 may be found, for example, in paragraph [0094]. Support for claim 118 may be found, for example, in paragraph [0078]. Support for claims 119 and 120 may be found, for example, in paragraph [0082]. Support for claim 121 may be found in the third full paragraph at page 3 (copy enclosed) of Provisional Application No. 60/170,140, which is incorporated by reference into the present application in its entirety (*see*, paragraph [0001] of US 2001/0031467). Support for claims 122 and 123 may be found, for example, in paragraph [0023]. Support for claim 124 may be found, for example, in paragraph [0019]. No new matter has been added.

Claim Rejections Under 35 U.S.C. § 102

Claims 1, 3-12, 15-18, 21, 39, 40, 42-60, 62-64, 68, 71-72, 75-81, 83-85, 89, 92, 93, 96, and 97 stand rejected under 35 U.S.C. § 102(b) as allegedly anticipated by Montforte *et al.* (WO 98/26095).

To facilitate allowance and without acquiescing to the rejection, Applicants have canceled the previously pending claims and submitted new claims 102-124. The new claims are directed to methods for separating a nucleic acid molecule of interest (*e.g.*, a genomic fragment of maternal origin) from a nearly identical nucleic acid molecule with a different parental origin (*e.g.*, a corresponding genomic fragment of paternal origin) in a population of nucleic acid molecules (*e.g.*, genomic DNA preparation from a biological sample). Such separation is performed based on the presence of a distinguishing element (*e.g.*, a single nucleotide polymorphism) in the nucleic acid molecule of interest, but not in the other nucleic acid molecule with a different parental origin. The presence of the distinguishing element in the nucleic acid molecule of interest allows a targeting element that comprises an oligonucleotide to form a perfect base pair match at or near the 3'-end of the oligonucleotide, which in turn allows a polymerase to extend the 3'-end of the oligonucleotide. The extension is performed in the presence of separation groups that comprise immobilizable and non-terminating nucleotides, which results in the attachment of separation groups to the targeting element. The resulting extension product together with the bound nucleic acid molecule of interest is then immobilized via the separation groups and separated from other nucleic acid molecules (including the corresponding nucleic acid molecule with a different parental origin) in the nucleic acid population that remains in the reaction mixture. Because the corresponding nucleic acid molecule with a different parental origin does not have the distinguishing element, it forms a base pair mismatch at or near the 3'-end of the oligonucleotide. Such a mismatch prevents the extension of the 3'-end of the oligonucleotide by the polymerase, and thus prevents the attachment of the separation groups to the targeting element. Accordingly, the nucleic acid molecule that is nearly identical to the nucleic acid molecule of interest but with a different parental origin is unable to be immobilized to a substrate that binds the separation groups.

Applicants respectfully submit that the Montforte *et al.* reference does not anticipate the currently claimed invention. First, the Montforte *et al.* reference fails to disclose a method for separating a nucleic acid of interest with one parental origin from the corresponding nucleic acid molecule with the other parental origin. There is no mention of haploid-specific nucleic acid isolation in the cited reference. Second, the Montforte *et al.* reference fails to disclose a distinguishing element that distinguishes a nucleic acid molecule of interest from a nucleic acid molecule that is nearly identical to the nucleic acid molecule of interest but with a different parental origin. The disclosure related to single nucleotide polymorphism detection in the cited reference is insufficient to teach the distinguishing element currently recited in the pending claims. Not all single nucleotide polymorphisms are capable of distinguishing a nucleic acid molecule of interest with one parental origin from the corresponding nucleic acid molecule with the other parental origin: Only those at heterozygous sites are. Third, the Montforte *et al.* reference fails to disclose a targeting element that comprises an oligonucleotide that (i) binds specifically to the target nucleic acid sequence of the nucleic acid molecule of interest, and (ii) overlaps the distinguishing element at or near the 3'-end of the oligonucleotide. Applicants disagree with the assertion in the Action regarding previously pending claim 58 that the cited reference "disclose[s] the method wherein the 3' terminus of the oligonucleotide is complementary to the polymorphism (*i.e.*, after extension, page 54, lines 5-7)." The specified location in the cited reference relates to the use of single nucleotide extension technique to detect single nucleotide polymorphisms. The oligonucleotide formed after extension in the cited reference would correspond to a targeting element with a separation group already attached to it, not to the targeting element itself, of the present application. If the above-noted assertion in the Action were correct, there would not be any disclosure of selective attachment of a separation group to a targeting element in the cited reference. Lastly, the Montforte *et al.* reference fails to disclose the selective attachment of a separation group only to the targeting element bound to the nucleic acid molecule of interest, but not to the targeting element bound to the other nucleic acid molecule that is nearly identical to the nucleic acid molecule of interest but with the other parental origin, in the presence of a polymerase.

In addition, Applicants respectfully traverse the rejections against previously pending claims 75 and 96 (which recite similar features as new claim 113). It is asserted in the Action that the Montforte *et al.* reference discloses the method wherein the nucleic acid molecule of interest is characterized (*i.e.*, following release of the mass label, the SNP is identified, Examples 6-8). This is incorrect. The molecule characterized in the cited reference corresponds to the extension product to which the nucleic acid molecule of interest is bound, not to the nucleic acid molecule of interest itself, of the present application.

Furthermore, Applicants respectfully traverse the rejections against previously pending claims 76 and 97 (which recite similar features as new claim 114). It is asserted that the Montforte *et al.* reference discloses the method wherein characterization identifies haplotype (*i.e.*, following release of the mass label, the SNP is identified, Examples 6-8). There is no mention of haploid-specific nucleic acid isolation in the cited reference. In addition, one SNP by itself does not constitute a haplotype, nor does a collection of SNPs alone. A haplotype is only generated by the linked association of SNP genotypes across a region of DNA. The cited reference only refers to genotypes, which cannot be considered a haplotype without knowing the linkage (*i.e.*, which alleles of the SNPs are from the maternal or paternal origin) across the SNPs.

In view of the above remarks, Applicants submit that the ground of rejection under 35 U.S.C. § 102(b) has been overcome. Withdrawal of this rejection is respectfully requested.

Claims 50-52 and 55 stand rejected under 35 U.S.C. § 102(e) as allegedly anticipated by Lundeberg *et al.* (U.S. Pat. No. 6,482,592).

To facilitate allowance and without acquiescing to the rejection in Action, Applicants have canceled the above-rejected claims. Accordingly, this ground of rejection under 35 U.S.C. § 102(e) is moot.

Claim Rejections Under 35 U.S.C. § 103(a)

Claims 1, 3-12, 15-18, 21, 39, 40, and 42-49 stand rejected under 35 U.S.C. § 103(a) as allegedly unpatentable over Ju *et al.* (U.S. Pat. No. 5,876,936) in view of Engelhardt *et al.* (U.S. Pat. No. 6,221,581).

Applicants respectfully submit that the cited references, either alone or in combination, fail to teach or suggest the subject matter as currently claimed in the present application. More specifically, the '936 patent relates to methods of enzymatic nucleic acid sequencing in which solid-phase capturable chain terminators are used. According to this cited reference, sequencing fragments are generated that comprise capturable chain terminators. The fragments are then captured on a solid phase and separated from the remaining components of the sequencing reaction. The fragments are then released from the solid phase, size separated and detected to yield sequencing data from which the sequence of the nucleic acid is determined. However, the '936 patent fails to teach or suggest any of the following features recited in the currently pending claims: (1) separating a nucleic acid molecule of interest with one parental origin from a nearly identical nucleic acid molecule with the other parental origin in a population of nucleic acid molecules, (2) the presence of a distinguishing element that distinguishes the nucleic acid molecule of interest from the other nucleic acid molecule that is nearly identical to the nucleic acid molecule of interest but with the other parental origin, (3) the presence of an oligonucleotide in a targeting element that binds specifically to the nucleic acid molecule of interest and overlaps the distinguishing element in the nucleic acid molecule of interest at or near the 3'-end of the oligonucleotide, and (4) selective attachment of a separation group only to the targeting element bound to the nucleic acid molecule of interest, but not to the targeting element bound to the other nucleic acid molecule that is nearly identical to the nucleic acid molecule of interest but with the other parental origin, in the presence of a polymerase.

The deficiencies in the '936 patent have not been remedied by the '581 patent. More specifically, the '581 patent is directed to methods for improving the binding of a series of consecutive nucleotide bases to a complementary target nucleic acid molecule in a sample and their use in isolating primer extension products. It fails to teach or suggest any of the above-noted features recited in the currently pending claims.

In view of the above remarks, Applicants submit that this ground of rejection under 35 U.S.C. § 103(a) has been overcome. Withdrawal of this rejection is respectfully requested.

Claims 53 and 54 stand rejected under 35 U.S.C. § 103(a) as allegedly unpatentable over Lundeberg *et al.* (U.S. Pat. No. 6,482,592).

To facilitate allowance and without acquiescing to the rejection in the Action, Applicants have canceled claims 53 and 54. Accordingly, this ground of rejection is moot.

Claims 61 and 82 stand rejected under 35 U.S.C. § 103(a) as allegedly unpatentable over Montforte *et al.* (WO 98/26095) in view of Snitman (U.S. Pat. No. 4,762,779).

Claims 65-67, 69, 86-88, and 90 stand rejected under 35 U.S.C. § 103(a) as allegedly unpatentable over Montforte *et al.* (WO 98/26095) in view of Beattie *et al.* (U.S. Pat. No. 6,268, 147).

Claims 70, 91, and 98-101 stand rejected under 35 U.S.C. § 103(a) as allegedly unpatentable over Montforte *et al.* (WO 98/26095) in view of Jones (U.S. Pat. No. 5,858,671).

Claims 73, 74, 94, and 95 stand rejected under 35 U.S.C. § 103(a) as allegedly unpatentable over Montforte *et al.* (WO 98/26095) in view of Radding *et al.* (U.S. Pat. No. 4,888,274).

Applicants respectfully submit that the cited references, either alone or in combination as indicated in the Action, do not teach or suggest the subject matter as currently claimed in the present application. More specifically, as discussed above, the Montforte *et al.* reference fails to disclose (1) a method for separating a nucleic acid of interest with one parental origin from the corresponding nucleic acid molecule with the other parental origin, (2) a distinguishing element that distinguishes a nucleic acid molecule of interest from a nucleic acid molecule that is nearly identical to the nucleic acid molecule of interest but with a different parental origin, (3) a targeting element that comprises an oligonucleotide that (i) binds specifically to the target nucleic acid sequence of the nucleic acid molecule of interest, and (ii) overlaps the distinguishing element at or near the 3'-end of the oligonucleotide, and (4) selective

attachment of a separation group only to the targeting element bound to the nucleic acid molecule of interest, but not to the targeting element bound to the other nucleic acid molecule that is nearly identical to the nucleic acid molecule of interest but with the other parental origin, in the presence of a polymerase.

The deficiencies in the Montforte *et al.* reference have not been remedied by either of the other cited references. More specifically, the '779 patent relates to a composition and a method for 5'-labelling polynucleotides undergoing solid phase synthesis. The '147 patent relates to a method for analyzing genomic DNA and expressed sequences using auxiliary oligonucleotides, preannealed to a single-stranded target nucleic acid to form a partially duplex target molecule. The use of auxiliary oligonucleotides is intended to improve the reliability of hybridization analyses and to avoid the inconvenient and costly labeling of numerous nucleic acid samples. The '671 patent relates to an iterative and regenerative method for sequencing DNA in intervals, starting at one end of a double stranded DNA segment. The '274 patent relates to a single-stranded nucleoprotein filament adapted to complex with a target duplex DNA having a selected base sequence. Each of these references fails to teach or suggest any of the above-noted features recited in the currently pending claims.

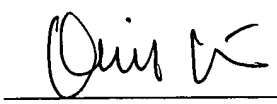
In view of the above remarks, Applicants submit that the aforementioned rejections under 35 U.S.C. § 103(a) have been overcome. Withdrawal of these rejections is respectfully requested.

The Director is authorized to charge any additional fees due by way of this Amendment, or credit any overpayment, to our Deposit Account No. 19-1090.

Applicants believe that all of the claims remaining in the application are now allowable. Favorable consideration and a Notice of Allowance are earnestly solicited.

Respectfully submitted,

SEED Intellectual Property Law Group PLLC



Qing Lin, Ph.D.
Registration No. 53,937

Enclosure: Page 3 of U.S. Provisional Application No. 60/170,140

QXL:kw

701 Fifth Avenue, Suite 6300
Seattle, Washington 98104-7092
Phone: (206) 622-4900
Fax: (206) 682-6031

(QXL:kw) 696809_2.DOC

solid support is desirable but limited to relatively short fragments. Magnetic bead extraction is a known effective way for retrieval of very long, genomic DNA but can similarly be performed for instance by centrifugation and density gradient separation using non-magnetic beads or particles (only DNA fragments containing a 'heavy handle' will precipitate while the other set will remain in the supernatant of the sample after centrifugation).

If the identity of the genotype used for fragment extraction is uncertain, the resulting genotyping of the remaining SNPs of the same sample will show if the extracted DNA was from one or both copies of the locus - in the first case the SNPs will all be of one kind (no heterozygotes).

The stability of the 'handle' for fragment extraction can be increased by several means, first of which is choosing the length of the primer such that the physical manipulation of the long DNA fragment does not result in a separation of the extended primer from the DNA. Selective extraction will be possible in certain casees by hybridization alone (= not using primer extension but by targeting significantly different patterns of one copy, thus selecting it during extraction over the other one with a higher melting temperature).

The extraction or immobilization of the selected fragments can be performed in 'bulk' for instance with magnetic beads or in very small quantities down to single molecules by using optically trapped beads for instance. It is easily possible to load many (> 100s) of strands onto one single bead, provided the concentration of DNA is sufficiently high. Amplification of the target DNA will usually be performed before the extraction step but it is conceivable that DNA can directly be extracted from fragmented genomic DNA. It is easy to work with long (> 100k base pair) stretches of genomic DNA, including at the single molecular level, which is desirable 1) to provide many SNPs at once rather than having to sequence or genotype several fragments that have to be mapped together and 2) if working with small tissue samples and for repeated analysis of variations within the same tissue.

The entire process can be automated, not only for use with magnetic beads (several systems exist) but also the process involving optically trapped beads or other methods as mentioned above. Simple and inexpensive microstructures can be made (see JD [1] and other publications) that serve as disposable yet resusable working and storage devices for the samples - amplified DNA would be loaded into a microstructure (for instance a glass - polymer hybrid structure) and preserved in there, extraction would be performed through a reversibly sealing port for instance by dipping a magnetized tip into the structure, or with a patch clamp pipette (for non-magnetic particles or fluid only - personal experience JD).

Recent progress in the design of miniaturized, handheld devices would make this a preferred platform in which the invention could be practiced in a variety of ways (using beads or other solid supports for strand separation or detection), especially if combined with a separate or integrated simple readout device such as a flatbed scanning device (fluorescent or other).

Other potential applications of the invention include assays that rely on the isolation or separate manipulation of nucleic acid sequences or entire chromosomes, in which a potentially small difference in sequence or structure is exploited to construct a specific 'handle' for the extraction of that motif. Applications would for instance include the transfection of genes between organisms, cloning, prenatal diagnostics in which a fertilized cell is to be analyzed for the presence of a certain sequence or marker,